

testing, respectively. The uteruses and decalcified femurs were then embedded in paraffin and sectioned for histopathological examination.

**Results:** Compared with the Sham group, the uterus indexes reduced significantly in OVX and ICT mice, with no significant difference between OVX and ICT mice. Histopathological examination of uterus showed a thinned endometrium with atrophic glands in OVX and ICT mice. Micro-CT analysis showed that the bone mineral density, bone volume/tissue volume, trabecular number and trabecular thickness of the 5th lumbar vertebrae all decreased significantly, and the trabecular separation increased in OVX mice. Mechanical testing showed that the biomechanical properties of the 5th lumbar vertebrae in OVX mice reduced, including the sectional elastic modulus, the maximum failure force and the energy absorbed until failure. Histopathological examination of femurs showed thin and spaced trabecular bone accompanied by increased bone marrow fat content in OVX mice. However, these osteoporotic phenotypes were rescued in ICT mice.

**Conclusion:** Icaritin can prevent OVX-induced osteoporosis in mice, without hyperplastic effect on uterus.

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##### INVESTIGATION OF HIERARCHICAL POROUS MICROSTRUCTURE OF SHANKBONE

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**Introduction and aims:** Bone possesses high fracture strength and toughness. Simultaneously, it is also relatively light. The excellent mechanical and physical properties of the bone are highly related to its elaborate microstructure refined by nature over many centuries. The detailed research on bone microstructure and the relationship between the microstructure and the properties of the bone can reveal the mechanism of strength and toughness as well as light-weight of the bone.

**Methods:** In this study, the hierarchical porous microstructure of a shank-bone was first analyzed by a scanning electron microscope (SEM) under different scales and directions. Then an image processing with a higher gray-scale resolution was used to analyze the SEM images of the porous microstructure of the bone under different scales. The porosity at different locations of the bone under different directions was identified. Lastly, the number and size of pores and density of the bone along different directions and under different scales were investigated by a MATLAB program.

**Results:** It was revealed that the bone was a porous bioceramics composite with particularly porous microstructure which varied by different scales, location and direction. It was indicated that the number and size of pores and density of the bone varied with the observed locations, directions and scales.

**Conclusion:** The polynomial expressions for the number and size of the pore and density of the bone are fitted from the relation curves between the number and size of the pores and density of the bone and the locations, directions and scales, which reveals the hierarchical porous microstructural characteristic of the bone.

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##### RELATIONSHIP BETWEEN MICROSTRUCTURE AND MECHANICAL BEHAVIOR OF CORTICAL BONE

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**Introduction and aims:** The mechanical behaviors of the bone, such as fracture susceptibility, are closely related to the microstructure of the bone. Therefore, the investigation on the relationship between the mechanical behavior and the microstructure of the bone is important. This investigation used the experiment and models of microscopic mechanics. The combination of the experiment and analytical models at microscopic scale could provide a fair understanding of the mechanical behavior of the bone.

**Methods:** In this work, the observation of scanning electron microscope, nanoindentation technique, hierarchical-model analysis and finite element computation at microscopic scale were used to investigate the relationship between the mechanical behavior of a cortical bone and its microstructure, such as its porosity, density and the direction of the hydroxyapatite-fiber sheets. In the hierarchical-model analysis, the osteons in the cortical bone were first considered as hollow fibers and the interstitial bone as matrix,

and then the osteons were further taken as a combination of many hydroxyapatite-fiber plies. Each fiber ply had different direction and modulus that provided different contributions to the mechanical properties of the bone. The porosity in the bone as well as the direction of the hydroxyapatite-fiber sheets in the osteons were modeled explicitly based on the images of the scanning electron microscope and X-ray diffraction. Computational results were obtained by applying uniform macroscopic stress to the boundaries of the microscopic model of the bone.

**Results:** The prediction of the macroscopic mechanical property corresponded reasonably well with the experimental data.

**Conclusion:** The relationship between the mechanical behavior and the microstructure of the bone is discussed in detail.

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##### TISSUE MINERAL DENSITY DEPENDENT MECHANICAL PROPERTIES OF INDIVIDUAL TRABECULA PLATES AND RODS DO NOT DIFFER IN ANATOMIC DIRECTIONS BUT INDIVIDUAL TRABECULAR DIRECTIONS

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**Introduction and aims:** Trabecular bone, susceptible to osteoporosis, consists of individual trabecular plates and rods, which are distributed distinctly along the longitudinal, transverse, or oblique anatomic directions of the skeleton (Fig.1). In each anatomic direction, mechanical properties of bone tissue are also expected to differ in (axial) or against (lateral) the direction of individual trabeculae, i.e., anisotropic mechanical properties. However, anisotropic mechanical properties of individual trabeculae along various anatomic directions are currently unclear. **Objectives** of this study were 1) to measure anisotropic tissue modulus and tissue mineral density (TMD) of individual trabeculae; 2) to examine their dependence on trabecular types and anatomic directions; and 3) to determine the relationship between anisotropic tissue modulus and TMD of individual trabeculae.

**Methods:** Twelve cylindrical human trabecular bone samples of proximal femurs were imaged with hydroxyapatite density calibration phantoms at 25 $\mu$ m resolution by micro-computed tomography ( $\mu$ CT). Individual trabecular types and their anatomic directions were determined using individual trabeculae segmentation (ITS) technique. On the embedded samples, micro-indentation tests were performed under wet condition on both axial and lateral cross-sections (Fig 1.C) of selected plates and rods in longitudinal (L), transverse (T), and oblique (O) directions, respectively (Fig 1.D). The point-by-point registered grayscale values of the  $\mu$ CT image at the indentation sites were converted to TMD using calibration phantoms.

**Results:** The tissue modulus and the co-localized TMD of trabecular plates were significantly higher than those of trabecular rods (Fig. 2A, B). The axial tissue modulus of individual trabeculae was significantly higher than the lateral tissue modulus (Fig. 2C). The tissue modulus correlated strongly with TMD. These correlations did not differ significantly between plates and rods or between different anatomic directions. However, the correlation of axial modulus was significantly different from that of lateral modulus (Fig. 2D).

**Conclusion:** We measured anisotropic elastic modulus of individual trabecular plates and rods of different anatomic directions. Surprisingly, the heterogeneous tissue modulus correlated with TMD similarly regardless of trabecular types and anatomic directions. The correlation only differs

